Available online at www.sciencedirect.com

ScienceDirect

Rice Science, 2016, 23(3): 119−131

Impact of Nitrogen, Phosphorus and Potassium on Brown Planthopper and Tolerance of Its Host Rice Plants

Md Mamunur RASHID[1], Mahbuba JAHAN[2], Khandakar Shariful ISLAM[2]

(*[1]Plant Physiology Division, Bangladesh Rice Research Institute, Gazipur, PO Box 1701, Bangladesh; [2]Department of Entomology, Bangladesh Agricultural University, Mymensingh, PO Box 2202, Bangladesh*)

Abstract: The brown planthopper (BPH), *Nilaparvata lugens* (Stål), appeared as a devastating pest of rice in Asia. Experiments were conducted to study the effects of three nutrients, nitrogen (N), phosphorus (P) and potassium (K), on BPH and its host rice plants. Biochemical constituents of BPH and rice plants with varying nutrient levels at different growth stages, and changes in relative water content (RWC) of rice plants were determined in the laboratory. Feeding of BPH and the tolerance of rice plants to BPH with different nutrient levels were determined in the nethouse. Concentrations of N and P were found much higher in the BPH body than in its host rice plants, and this elemental mismatch is an inherent constraint on meeting nutritional requirements of BPH. Nitrogen was found as a more limiting element for BPH than other nutrients in rice plants. Application of N fertilizers to the rice plants increased the N concentrations both in rice plants and BPH while application of P and K fertilizers increased their concentrations in plant tissues only but not in BPH. Nitrogen application also increased the level of soluble proteins and decreased silicon content in rice plants, which resulted in increased feeding of BPH with sharp reduction of RWC in rice plants ultimately caused susceptible to the pest. P fertilization increased the concentration of P in rice plant tissues but not changed N, K, Si, free sugar and soluble protein contents, which indicated little importance of P to the feeding of BPH and tolerance of plant against BPH. K fertilization increased K content but reduced N, Si, free sugar and soluble protein contents in the plant tissues which resulted in the minimum reduction of RWC in rice plants after BPH feeding, thereby contributed to higher tolerance of rice plants to brown planthopper.

Key words: *Nilaparvata lugens*; relative water content; host tolerance; nitrogen; phosphorus; potassium; rice; nutrient subsidy

The brown planthopper (BPH), *Nilaparvata lugens* (Stål) (Homoptera: Delphacidae), is a major insect pest of rice in Asian countries, causing heavy crop damage through 'hopper burn' (Bottrell and Schoenly, 2012; Ali et al, 2014). Nutrition management is one of the most important practices for high production system, but it may affect on host-insect interaction. Insect behavior and life parameters are affected by environmental factors, such as temperature, moisture, habitat morphological and chemical components of host plants, especially by the nutrients, such as nitrogen, sugars, amino acids and semio-chemicals in host plants (Fischer and Fiedler, 2000), and plant water content is another important factor (Slansky and Rodriguez, 1987). Several studies have indicated the importance of host plant quality on herbivorous insects (Awmack and Leather, 2002; Bado et al, 2002). Abiotic heterogeneity through crop nutrition can affect the susceptibility of plants to insect pests by altering plant tissue nutrient levels (Altieri and Nicholls, 2003) and morphology (Moon and Stiling, 2000). Excessive and/or inappropriate use of inorganic fertilizers can

---

Received: 11 May 2015; **Accepted:** 26 August 2015

Corresponding author: Md. Mamunur RASHID (mrashidbrri@gmail.com)

Copyright © 2016, China National Rice Research Institute. Hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Peer review under responsibility of China National Rice Research Institute

http://dx.doi.org/10.1016/j.rsci.2016.04.001

cause nutrient imbalances and lower pest resistance (Altieri and Nicholls, 2003; Marazzi et al, 2004). Outbreaks of planthopper populations are sensitive barometers of crop mismanagement (Sogawa et al, 2009).

Nitrogen (N) content is regarded as an indicator of plant quality and also one of the most important performance limiting factors of herbivores (Lu and Heong, 2009). N application is reported to induce succulence in rice plants which makes them more prone to insect pests (Salim, 2002b). Heavy applications of N fertilizer may not affect insect biology directly but change the host-plant morphology, biochemistry and physiology, which can improve nutritional conditions for herbivores (Bernays, 1990), thus playing a key role in modifying and reducing host resistance to them (Barbour et al, 1991). N limitation is well documented in insect herbivores, but phosphorus (P) limitation is poorly studied (Huberty and Denno, 2006), although insect herbivores require not only N but also P to synthesize their proteins (Sterner and Elser, 2002). P is required for ATP and nucleic acid synthesis (RNA and DNA), and thus protein production (Sterner and Elser, 2002). As a result, its limitation can impose severe consequences for cellular function and ultimately the growth rate of consumers (Elser et al, 2000; Sterner and Elser, 2002). P limitation has been documented widely in many species of aquatic invertebrates, particularly in *Daphnia* (Elser et al, 2001). Potassium (K) has a critical role in plant physiology (Wyn Jones, 1999). In rice, K provides regulatory control over different processes like transpiration, starch synthesis, sucrose translocation, respiration and lipid synthesis (Tisdale et al, 1985). K nutrition has a profound effect on the profile and distribution of the primary metabolites in plant tissues. Changes in metabolite concentrations induced by K are multiple and it includes K dependence of metabolic enzymes, photosynthesis and long-distance transport. The primary metabolites such as soluble sugars particularly reducing sugars, organic acids and amino acids tend to increase in K deficient plants (Amtmann et al, 2008). Approximately 60 enzymes have been shown to depend on K *in vitro* for their activities, many of which are involved in sugar or N metabolism (Wyn Jones and Pollard, 1983). It is also important to note that a direct effect of K nutrition on enzyme activity occur only if cytoplasmic K concentrations are changed (Amtmann et al, 2008). Because of efficient cellular K homeostasis (Walker et al, 1996), a decrease of cytoplasmic K concentration can be expected to occur only during prolonged K deficiency (Wyn Jones, 1999). It was reported that plant damage by insect is comparatively less in K applied plants due to reduced carbohydrate accumulation, elimination of amino acids (Baskaran et al, 1985), higher silica content and increase in the sclerenchymous layer (Dale, 1988). The mechanism of rice plants damaged by BPH through hopper burn is similar with that of drought stress which is closely related to the relative water content (RWC) of plants. Typical RWC in rice plants at wilting is in the range of 60% to 70% (Lu et al, 2004).

Host plant tolerance to BPH is an important issue for management of BPH. A little information is available on the effects of N, P and K on biochemical composition of BPH and its host rice plants. Information on the relationships between biochemical compositions of rice plants and its tolerance to BPH particularly in reference to different nutritional status is also very limited. We studied the effects of three major plant nutrients i.e. N, P and K on the biochemical compositions of both BPH and its host rice plants, feeding of the insect, fluctuations of RWC in plants, and tolerance of plants to the pest BPH.

MATERIALS AND METHODS

Preparation of soil and host plant establishment

The potted plants of different nutrient levels were prepared separately for the experiments at nethouse of Bangladesh Rice Research Institute, Gazipur, Bangladesh. Pot soil was fertilized with three levels of each nutrient: N (0, 100 and 200 kg/hm^2), P (0, 20 and 40 kg/hm^2) and K (0, 60 and 120 kg/hm^2) and combination of all the levels. The soil was slightly acidic (pH 5.3), low organic carbon (0.72%) and deficient in N, P and K, where levels of all the three nutrients were below the critical level. Approxiamately 2 kg dry soil was taken in each pot with 16 cm height and 14 cm diameter. The exact amount of input fertilizers was calculated based on the amount of soil in each pot. Fifteen-day-old rice seedlings were transplanted (3 hills per pot and 2 seedlings per hill) in each pot. The rice variety BR3 was selected because of its year round growing habit and susceptibility to BPH. To obtain continuous supply of host plant materials for each BPH culture, the experiment seedlings were transplanted at a 15 d interval. Plants with different nutrient treatments were labeled and placed under natural condition in nethouse. All the factorial pot experiments with 27 treatment combinations were laid out in a completely randomized design and replicated four times considering each pot as a replicate.

Effects of nutrient subsidies on biochemical constituents of rice plants and BPH

Effects of N, P and K treatments on biochemical compositions of rice plants were determined on plants that were never exposed to BPH. The effects on rice plants at four stages, namely mid-tillering, panicle initiation, heading and maturity (soft dough) stages, were determined from four randomly chosen pots for each treatment. Four plants were taken randomly from each pot and oven-dried at 70 °C ± 5 °C for 72 h. Plant samples were then ground in a Wiley Mill (Thomas Scientific Inc., USA) and analyzed for contents of N, P, K, silicon (Si), total free sugars and soluble proteins. For analysis of total N, 200 mg ground sample was digested in a micro Kjeldahl flask with the same weight of catalyst mixture (K_2SO_4:$CuSO_4$:Selenium, 50:10:1) and 3 mL of concentrated H_2SO_4 until clear, cooled and brought up to the volume with distilled water. N was determined volumetrically by the micro Kjeldahl distillation method (Yoshida et al, 1976). For analysis of P and Si concentrations, 1 g sample was digested with 10 mL acid mixture of $HClO_4$, HNO_3 and H_2SO_4 (300:750:150) on a hot plate at 500 °C until a gelatinous white residue remained to which distilled water was added after cooling. The mixture was filtered through Whatman #42 filter paper. P concentration was determined colorimetrically through a Spectro Photometer (V-630, JASCO, Japan) at 420 nm wavelength. The residue on the filter paper was burnt at 500 °C for 4 to 5 h for gravimetric determination of crude Si concentration. For analysis of K concentration, 1 g plant sample was soaked in 25 mL of 1 mol/L HCl for 24 h and filtered through Whatman #43 filter paper. K concentration was analyzed in the filtrate using a Flame Photometer (410 FP, Sherwood, UK) at 214 nm wavelength. For estimation of soluble protein and total free sugar concentrations, plants were analyzed based on dry weight (Lowry et al, 1951; Yoshida et al, 1976). Brown planthopper population required for the experiments were obtained from a mass culture raised on rice plants with native nutrients maintained in nethouse. A total of 200 the first instar nymphs of BPH were released on 30-day-old potted rice plants established with different nutrient treatments. For determination of the nutrient content of BPH, four samples each with 25 emerging adult planthoppers of each treatment was collected and oven-dried prior to analysis. They were ground in a Wiley Mill (Thomas Scientific Inc., USA) and analyzed for contents of N, P and K. The grand mean of all experimental treatment plants and BPH were used to assess the overall chemical contents of rice plants and BPH. Mismatch in N, P and K contents between rice plant and BPH was compared.

Effects of nutrient subsidies on honeydew secretion

Amount of feeding BPH was measured indirectly by the quantity of honeydew secretion. The honeydew excreted by BPH reared on rice plants with different nutrient treatments were collected in a feeding chamber following the method developed by Pathak and Heinrichs (1982) with partial modification. The feeding chamber consisted of an inverted transparent plastic cup resembling a conical funnel and placed over a filter paper treated with 0.2% Bromocresol green solution. The whole apparatus was rested on a plastic petridish. Adult females with uniform age and size were collected from the mass culture and starved for 3 h. The leaf sheaths of the plants in each treatment were wiped well with tissue paper to remove any kind of moisture on the surface. Five 3-day-old BPH adult females were placed into the conical chamber through a hole at the top of the cup. A cotton-wad was then placed in the hole to prevent from escaping of the insects. The BPH was allowed to feed for 48 h and then the filter paper was removed. Areas of the filter paper receiving the honeydew turned purple bluish. The spots were marked on tracing paper and the feeding areas were measured by counting the squares in mm^2 girded transparent graph paper.

Effects of nutrient subsidies on tolerance of rice plants to brown planthopper

The changes of RWC in rice plants and the severity of plant damage due to BPH feeding were determined. A total of 100 the second instar nymphs were introduced into mylar cage containing 35-day-old potted rice plants trimmed to 6 tillers for all the 27 treatments. The RWC of each treatment was measured twice, first before infestation and then at 5 d after release of BPH on rice plants, and the reduction of RWC in plants due to BPH feeding was calculated. To determine RWC in rice plants, four samples were collected from each treatment. The top most fully expand leaves were sampled and the mid leaf section of about 5–10 cm^2 was cut with scissors and then subsequently placed in a pre-weighed airtight vial to obtain leaf sample weight (W). The leaf samples were immediately hydrated by distilled water up to full turgidity in the refrigerator at about 10 $^{\circ}$C for 4 h. They were then taken out of water, well dried with filter paper and weighed to obtain the fully turgid weight (TW). Samples were then dried at 80 $^{\circ}$C for 24 h and weighed to calculate the dry weight (DW). Relative water content was computed as:

$$RWC\ (\%) = [(W - DW) / (TW - DW)] \times 100.$$

Damage to the rice plants was rated daily based on the standard evaluation system for rice (IRRI, 1980) as follows: 0, no damage; 1, very slight damage; 3, partially yellowing of the first and second leaves of most plants; 5, pronounced yellowing and stunting or wilting of 10% to 25% of the plants; 7, wilting or dead of more than half of the plants and severely stunting or dying of the remaining plants; 9, death of all plants.

The survival duration of rice plants or the duration from infestation with BPH to the death of rice plants was recorded.

Statistical analysis

Analysis of variance (ANOVA) was performed using Crop Stat 7.2 (IRRI, 2007). The mean differences among the treatments were compared by multiple comparison tests using the Duncan's multiple range test (DMRT). A statistic paired sample t-test was used to assess the mismatch in N, P and K stiochiometry between rice plants and BPH (SPSS, 2007). Regression analysis was conducted using Crop Stat 7.2 (IRRI, 2007) to examine the effect of independent variables plant tissue nutrients i.e. N, P, K, Si, total free sugars and soluble proteins on the dependent variables, honeydew secretion by BPH and plant tolerance to the pest.

RESULTS

Effects of nutrient subsidies on biochemical compositions of rice plants and BPH

Nitrogen fertilizer application to the rice plants resulted in a significant increase in N content ($P < 0.001$) in both rice plants and BPH (Table 1 and Fig. 1-A). Application of K decreased N content in rice plants significantly ($P < 0.001$) but not in BPH ($P = 0.069$) (Table 1 and Fig. 1-C). However, P application did not significantly change N content of both rice plants ($P = 0.078$) and BPH ($P = 0.14$) (Table 1 and Fig. 1-B). Fertilizing with P significantly increased the P content in rice plant tissues ($P < 0.001$) but not in BPH ($P = 0.068$) (Table 1 and Fig. 1-E). Different levels of N and K subsidies to rice plants did not alter P content in both rice plants and BPH (Table 1 and Fig. 1). Increase of K application significantly increased K content in rice plants ($P < 0.001$), but K content remained unchanged in BPH ($P = 0.172$) (Table 1 and Fig. 1-I). Different levels of N and P application to rice plants did not alter K content in both the organisms ($P > 0.05$, Table 1 and Fig. 1). Application of the nutrients N and K was found to decrease the Si content in rice plants significantly ($P < 0.001$, Table 1 and Fig. 2). This decreasing rate of Si was more than 50% with N application but it was about 11% with K. The amount of Si remained unaffected with the application of P fertilizer to rice plants ($P = 0.133$, Table 1 and Fig. 2-B). No significant change in total free sugar content in rice plants was found due to application of N ($P = 0.477$) and P ($P = 0.885$), but it caused a significant reduction with the application of K ($P < 0.001$) (Table 1 and Fig. 2). There was an increase in soluble protein content in rice plants with increasing levels of N ($P < 0.001$, Fig.

Table 1. Analysis of variance for biochemical compositions in rice plants and brown planthopper (BPH) affected by nitrogen (N), phosphorus (P) and potassium (K) subsidies (*F* values).

Source of variance	*df*	Nitrogen		Phosphorus		Potassium		Silicon in rice	Total free sugar in rice	Soluble protein in rice
		Rice	BPH	Rice	BPH	Rice	BPH			
N	2	2166.51**	12.87**	2.45	0.16	0.11	0.01	2 486.75**	0.73	3 300.13**
P	2	2.58	2.04	544.36**	2.77	0.45	0.15	2.33	0.12	0.72
K	2	392.74**	2.80	2.08	0.50	440.21**	1.77	33.03**	573.66**	822.94**
N × P	4	2.05	0.25	2.44	0.19	0.20	0.70	0.87	0.29	0.17
N × K	4	53.12**	0.85	0.94	0.56	0.03	0.92	3.73**	0.61	15.65**
P × K	4	2.21	1.04	2.28	0.74	0.14	0.67	0.75	0.07	0.21
N × P × K	8	1.40	0.60	1.26	0.58	0.17	1.69	0.86	0.10	0.29
Error	81									
CV (%)		2.65	3.22	4.18	4.45	2.92	1.75	5.33	1.98	2.48

**, Significant at the 1% level of probability.

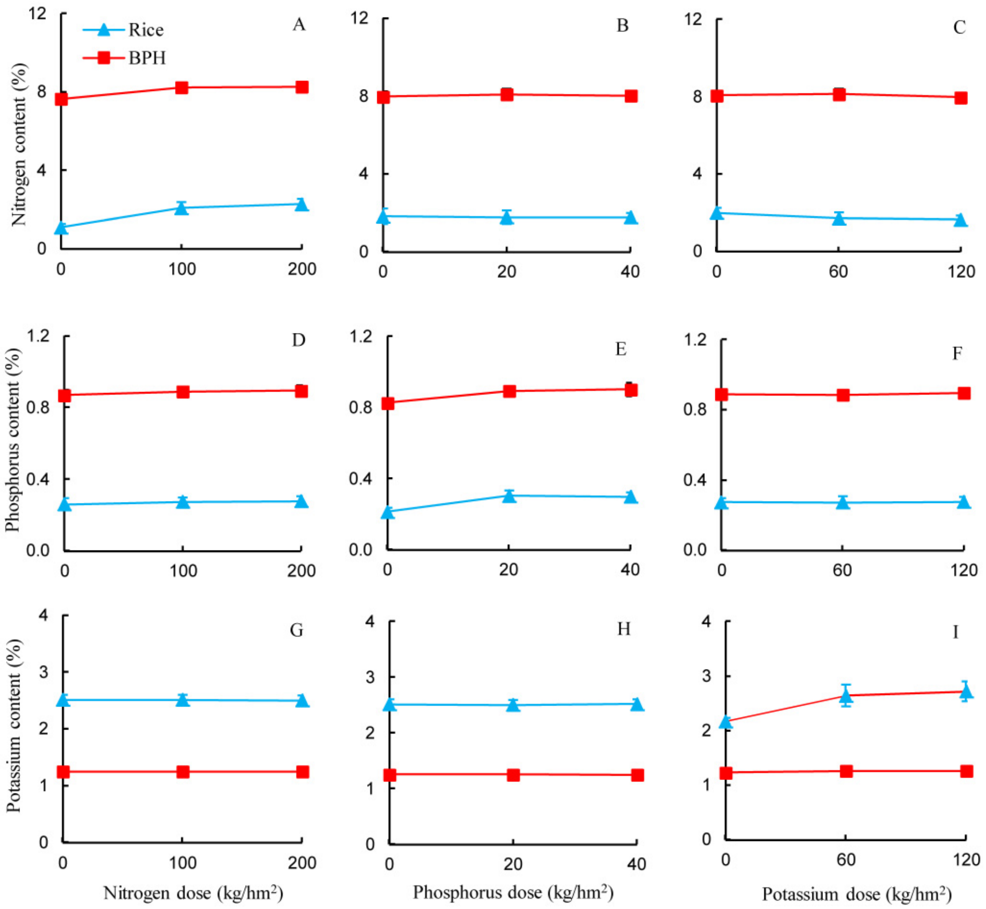

Fig. 1. Response of nutrient levels on nutrient contents of brown planthopper (BPH) and its host rice plants.
Bar indicates the standard error (*SE*).

2-A), but no change was recorded with the addition of P ($P = 0.494$, Fig. 2-B). However, the concentration of soluble proteins in rice plant tissues decreased significantly with the application of K ($P < 0.001$, Table 1 and Fig. 2-C). The interaction between applied N and K was significant for concentration of N ($P < 0.001$), Si ($P = 0.017$) and soluble proteins ($P < 0.001$) in the tissue of rice plants.

Mismatch in N, P and K content between rice plants and brown planthopper

Nutrient content varied significantly between BPH and its host rice plants as shown in Fig. 3. Concentrations of N and P in the body of BPH were significantly higher than those in rice plant tissues at all the four growth stages, mid-tillering, panicle initiation,

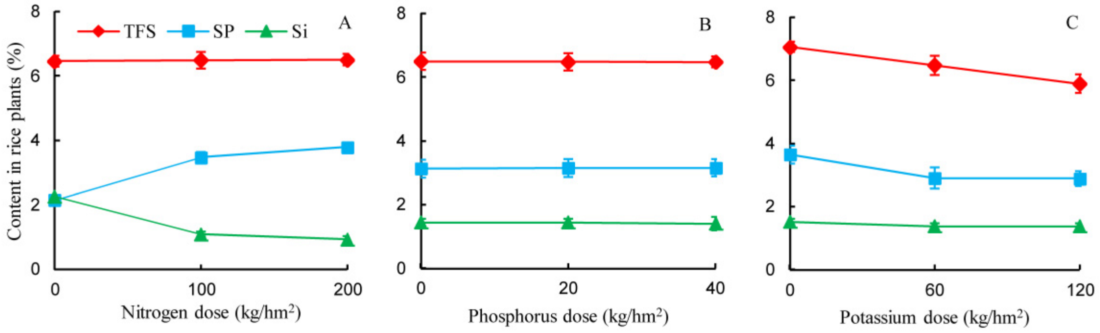

Fig. 2. Response of nutrient levels on silicon (Si), total free sugar (TFS) and soluble protein (SP) contents in rice plants.
Bar indicates the standard error (*SE*).

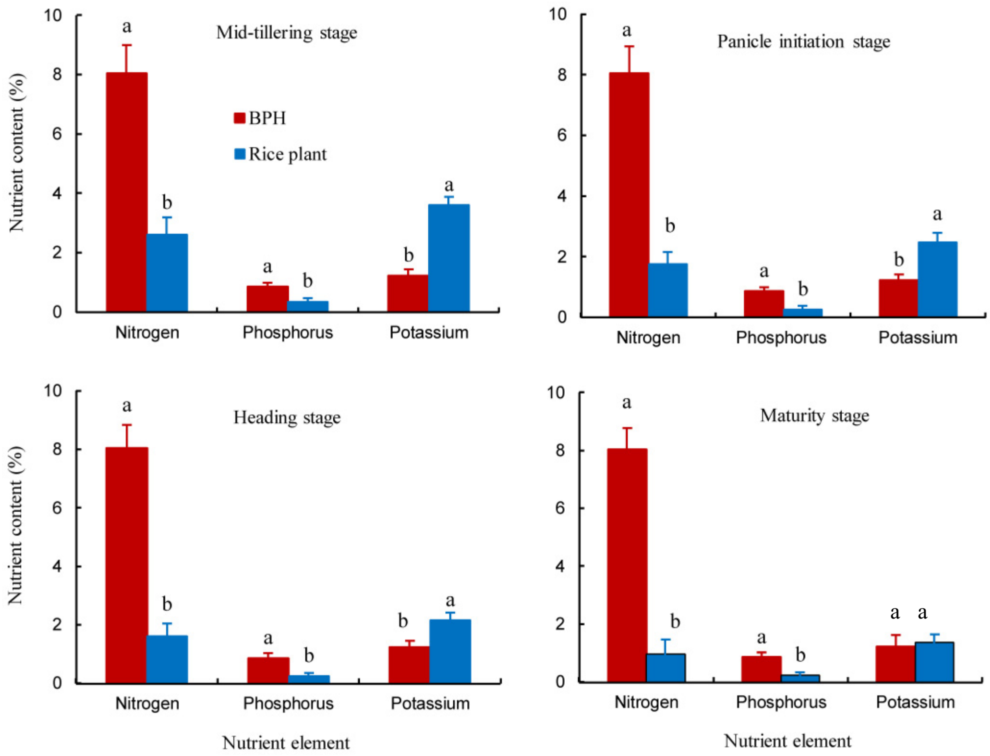

Fig. 3. Status of elemental contents in brown planthopper (BPH) and rice plants at different stages.
Bar indicates the standard error (*SE*).

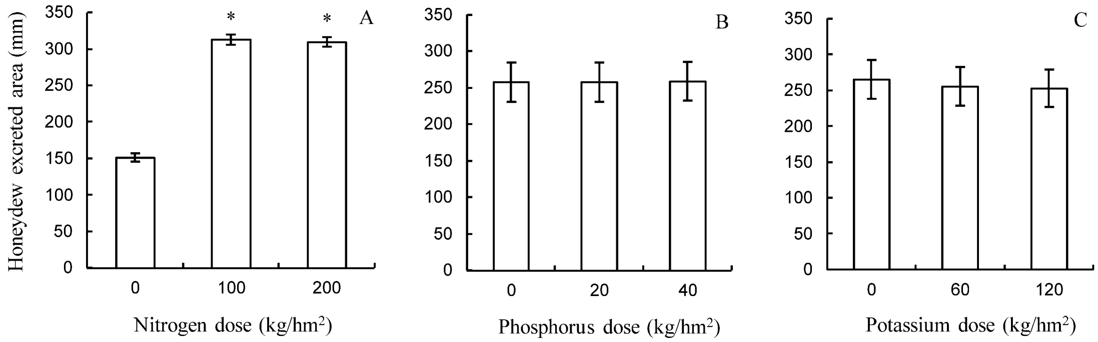

Fig. 4. Feeding of brown planthopper (BPH) measured indirectly by honeydew excretion in different nutrient treated rice plants.
Bar indicates the standard error (*SE*).

heading and maturity stages ($P < 0.001$, Table 2). The concentration of N in BPH was around four times higher than that of rice plants. Similarly, the P content of BPH was also around three times higher than that of the plants. In contrast, the concentration of K had an opposite trend and it was significantly higher in rice plant tissues at mid-tillering, panicle initiation and heading stages than that of BPH ($P < 0.001$, Table 2). However, K concentration did not differ significantly between BPH and rice plants at the maturity stage of the plants ($P = 0.057$, Table 2).

Table 2. Mean comparison results for nitrogen (N), phosphorus (P) and potassium (K) contents between brown planthopper (BPH) and its host rice plants (*t* value).

Rice stage	*df*	N content	P content	K content
Mid tillering	26	150.32**	111.80**	80.90**
Panicle initiation	26	106.79**	134.16**	21.95**
Heading	26	163.32**	136.40**	162.92**
Maturity	26	163.72**	108.38**	1.94

**, Significant at the 1% level.

Effects of nutrient subsidies on honeydew excretion

Effects of varying levels of N, P and K on honeydew secretion by BPH are presented in Fig. 4. Amount of

Table 3. Analysis of variance for honeydew secretion, changes in relative water content (RWC) and complete damage duration of rice plants feeding brown planthopper (BPH) as affected by nitrogen (N), phosphourus (P) and potassium (K) subsidies (*F* value).

Source of variation	*df*	Honeydew secretion	RWC of BPH free plants	RWC at 5 d after infestation by BPH	Reduction of RWC	9th grade plant damage duration
N	2	601.80**	274.53**	1 338.95**	1 385.84**	242.70**
P	2	0.04	9.36**	0.55	0.56	0.66
K	2	2.74	37.75**	17.56**	16.46**	19.70**
N × P	4	0.08	0.82	0.18	0.17	0.69
N × K	4	0.14	4.55**	11.48**	10.41**	2.62*
P × K	4	0.06	0.72	1.45	1.44	1.43
N × P × K	8	0.08	1.24	1.46	1.39	1.01
Error	81					
CV (%)		4.78	0.78	1.25	2.43	8.54

* and **, Significant at the 5% and 1% levels of probability, respectively.

honeydew excretion increased significantly with N application to rice plants ($P < 0.001$, Table 3). However, it did not increase linearly with the N application rate. The highest amount of honeydew secretion was recorded from the individuals feed on plants treated with nitrogen level at 100 kg/hm^2 followed by the levels at 200 and 0 kg/hm^2. Honeydew secretion by BPH did not differ significantly among different P treatment levels ($P = 0.959$, Table 3). Increasing amount of K fertilizer application to rice plants promoted to a slight decrease in the honeydew excretion by BPH, but it was statistically insignificant ($P = 0.07$, Table 3). Analysis of variance did not detect significant effect from the interactions of the three nutrients (N, P and K) for honeydew secretion by BPH ($P > 0.05$, Table 3).

Effects of nutrient subsidies on tolerance of rice plants to brown planthopper

Effects of different levels of N, P and K on RWC of BPH unfed and fed rice plants are presented in Fig. 5. RWC in BPH free rice plants increased significantly with the application of all the three nutrients N, P and K ($P < 0.001$, Table 3). Reduction rate of RWC in rice plants due to BPH feeding differed significantly among different nutrient treatments. It decreased significantly at higher rate in N applied plants than without N application ($P < 0.001$, Table 3). Reduction of RWC by BPH infestation was significantly less in plants fertilized with any level of K than in plant without K application ($P < 0.001$) and interaction

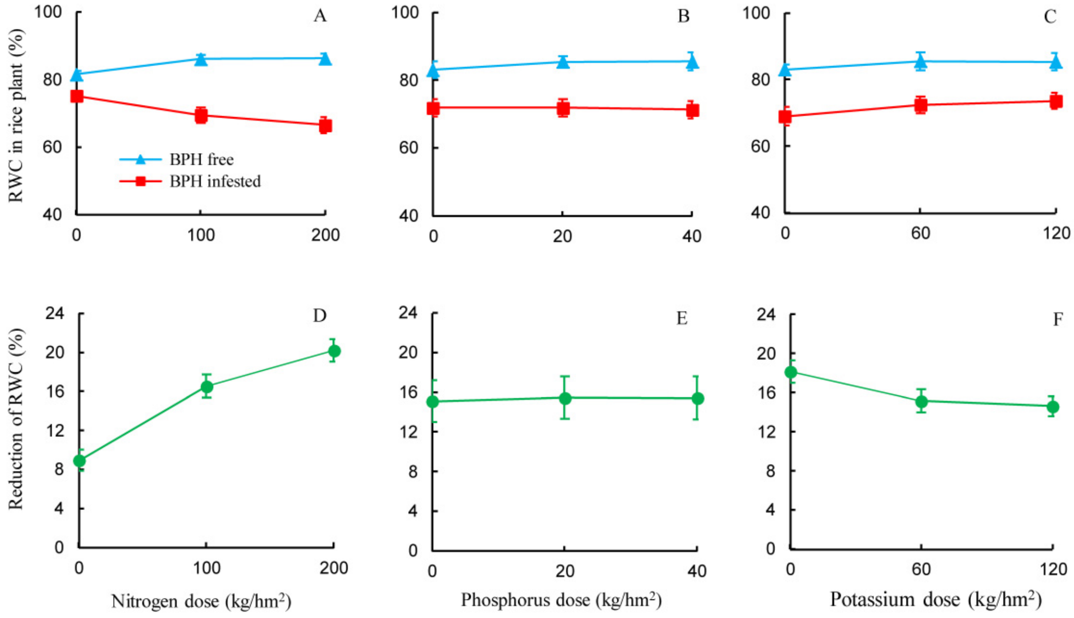

Fig. 5. Relative water content (RWC) and reduction of RWC due to brown planthopper (BPH) feeding after 5 d in different nutrient treated rice plants after release of BPH.

Bar indicates the standard error (*SE*).

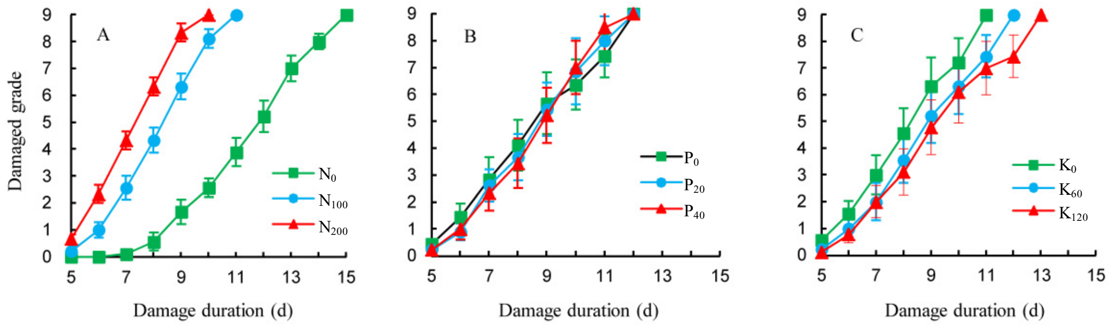

Fig. 6. Dynamics of damage grade of rice plants by brown planthopper (BPH) as affected by different levels of nutrient application.
Bar indicates the standard error (*SE*).

between N and K was significant ($P < 0.001$, Table 3). However, there was no significant difference among P fertilization treatments in changing RWC of rice plants damaged by BPH ($P = 0.587$, Table 3). Interaction effects of N and P, P and K and all the three nutrients (N, P and K) were insignificant for reduction of RWC of rice plants by BPH feeding ($P > 0.05$, Table 3).

Complete damage (9th grade) durations by BPH injury varied significantly among rice plants fertilized with different nutrient treatments and are presented in Fig. 6. The duration from infestation with the second instar BPH nymphs to the death of rice plants declined significantly with the increase of N application rate ($P < 0.001$, Table 3). While, it required significantly more duration to cause complete damage when K was applied to the plants ($P < 0.001$) and interaction between N and K was significant ($P = 0.047$) (Table 3). Complete plant damage duration (9th grade) remained unaffected with whatever the level of P was applied ($P = 0.821$, Table 3). No significant difference was evident in the duration for complete plant damage by BPH when the interactions of N and P, P and K and all the three nutrients (N, P and K) were taken into consideration ($P > 0.05$, Table 3).

Relationships between life parameters of BPH and chemical contents in rice plants

All the relationships of dependent variables (honeydew secretion by BPH and rice plant tolerance to the pest)

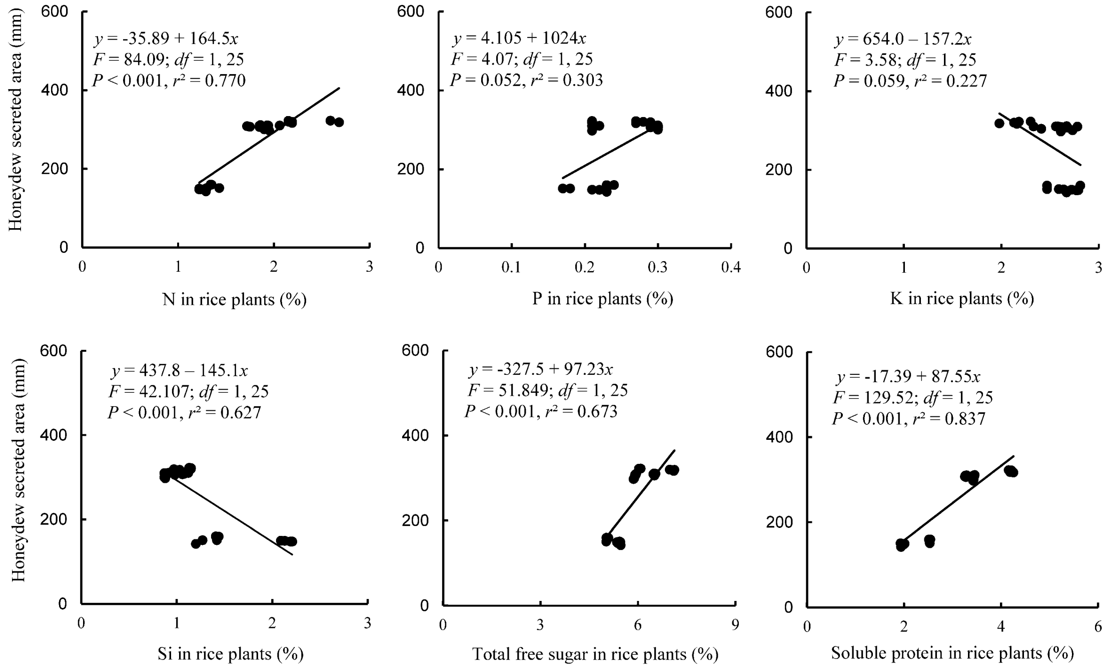

Fig. 7. Relationship between brown planthopper (BPH) feeding and different nutrients nitrogen (N), phosphorus (P), potassium (K), silicon (Si), total free sugar and soluble proteins contents in rice plants.

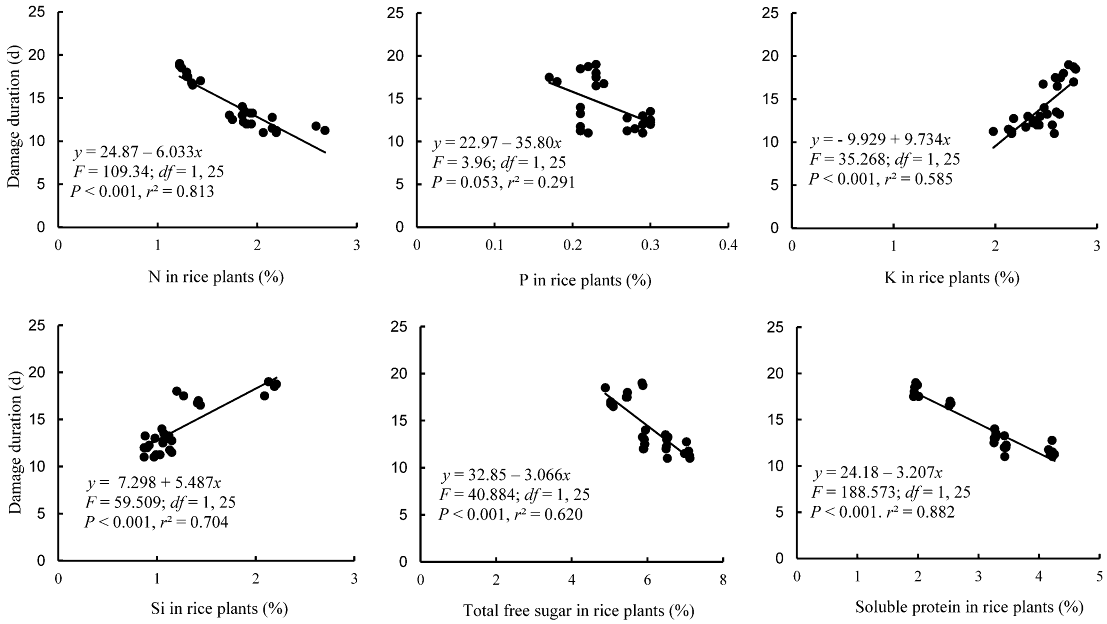

Fig. 8. Relationships between nitrogen (N), phosphorus (P), potassium (K), silicon (Si), total free sugar and soluble protein contents in rice plants and its 9th grade damage by brown planthopper (BPH).

with independent vatiables plant tissue nutrients (N, P, K, Si, total free sugars and soluble proteins) are summarized in Figs. 7 and 8. Relationships between honeydew secretion and plant tissue nutrients N, Si, total free sugars and soluble proteins fitted the linear model. Honeydew secretion positively correlated ($P < 0.001$) with concentrations of N, total free sugar and soluble protein but negatively with Si in rice plant tissues. However, linear but weak relationship was observed between honeydew secretion by BPH and nutrients concentration of P (P = 0.052) or K (P = 0.059) in rice plant tissues. Relationship between complete damage duration of rice plants due to BPH feeding and N, K, Si, total free sugar and soluble protein contents in rice plant tissues also showed linear model. Complete plant damage duration was negatively correlated with plant tissue N, total free sugar and soluble protein contents in rice plant tissues ($P < 0.001$), while positively correlated with K and Si contents in rice plant tissues. However, relationship between damage duration of rice plants and its P content was also linear but weak (P = 0.053).

DISCUSSION

N, P and K are essential macronutrients of rice that must often be applied in rice fields through different forms of fertilizers to maintain the productivity of cropped soil and prevent deficiencies of these essential nutrients from limiting crop yields. Three levels of N, P and K were used in this study. The ranges of N, P and K were chosen to bracket those used in previous laboratory experiments and to include the spectrum of nutrient contents that has occured in cultivated rice fields in Bangladesh. Usually, for rice cultivation, the nutrients N, P and K are recommended at the rates of 100, 20 and 60 kg/hm^2, respectively, but sometimes some of the farmers exceed the usual level with the expectation to obtain higher yield. In many cases, the increase of the nutrients by them is not proportionate. Application of over doses of N is frequently common and this can be a reason for the BPH outbreak in rice fields. Considering this context, the doses of the present experiment were selected to determine the consequences of variable amount of N, P and K as subsidies to the plants and its pest BPH. In general, enhancement of nutrient subsidies results in higher nutrient content in rice. Increased supply of inorganic fertilizers enhances nutrient uptake through their roots. In the present study, increasing levels of N application to rice plants resulted in a significant increase in N content in plant tissues. Moreover, it also increased soluble protein content and decreased Si content in the leaf sheath of rice plants, which might play a vital role in BPH feeding and host plant tolerance. A report indicates that supplementation of N had similar effect and also found to decrease the level of Zn in rice plants (Salim, 2002b). P content in rice plant tissue was found higher due to application of P and the maximum amount of P was recorded when the plant was treated with 20 mg/kg P, which is in agreement with the reports of Subbaiah (1991) and Mongia et al (1998). Application of K fertilizer to the rice plants increased the concentration of K in plant tissues, but it decreased the contents of N, Si, soluble protein and total free sugar. It was reported that increase in the application of K in the culture solution increased K content, but decreased N, Si and soluble protein contents in rice plants (Salim, 2002a). Other reports showed K fertilization seemed to increase K level (Stiling and Moon, 2005; Sarwar, 2012) and decreased N level (Clark, 1982) in plant tissues. Experimental results clearly indicated that increasing application rate of N fertilizer to rice plants have significantly increased the N content of BPH but the levels of P and K remained unchanged. Fertilization with P and K to rice plants did not significantly alter the N, P and K contents in the body of BPH.

Study on nutrient mismatch between the plant and insect is quite important for studying the feeding aspects of insects. The difference in the particular nutrient level in host plants and harbivour will play a vital role in causing damage to the plant. Some reports showed that stoichiometric mismatches between the insects' requirements and their diet play an important role on the insect body elemental concentrations and life history traits (Elser et al, 2000; Huberty and Denno, 2006; Visanuvimol and Bertram, 2011). In the present study, the concentrations of N and P in rice plants were found remarkably low compared to those in BPH bodies. This mismatch highlighted the poor nutrient quality of rice plants for BPH. Many authors reported that the concentrations of mineral nutrients are lower in plants than in herbivores (Elser et al, 2000), and specially, phytophagous insects have much higher N and P contents than their host plants (Elser et al, 2000; Fagan et al, 2002; Visanuvimol and Bertram, 2011). Insects with relatively high body nutrient concentrations require nutrient-rich foods otherwise they would suffer reduced growth. As a consequence, insects choose foods that match their stiochiometric requirements (Jensen et al, 2006). Our study clearly showed that BPH body contained very high amount of N, which indicated that requirement of this particular element is always demanding, as the host plants normally supplied insufficient amount of N. These might be the important reasons why most herbivores feed more on host plants with high N content. Although the concentration of P in rice plants is also lower than BPH, P fertilization can not cause any significant change in feeding of the pest probably for its lower requirements than N. However, it is quite difficult to draw a complete conclusion about the mismatch of P between the BPH body and rice plant tissues, which demands further investigation. Concentration of K in rice plant tissues was higher than that of the BPH bodies, suggesting that rice plant itself is a good food source of K for BPH at anywhere grown at any level.

Feeding of BPH is usually determined indirectly by measuring the amount of honeydew secretion. In the present study, honeydew excretion by BPH increased with the increase of the N level but not with P and K levels. Nitrogen application to the rice plants up to 100 kg/hm^2 resulted in the increase of honeydew excretion. This might be for the higher preference of BPH to suck more on plants fertilized with 100 kg/hm^2 N containing higher concentrations of N and soluble protein and lower Si concentration than those of plants without N fertilization. However, further increase of the N dose to 200 kg/hm^2 reduced honeydew excretion by BPH. This might be due to BPH decrease food consumption on better quality food resources, probably as it had reached the saturation point earlier. There are many similar reports that the rice plants supplied with N fertilizer have high feeding rates and honeydew excretion (Lu et al, 2005; Prasad et al, 2005; Lu and Heong, 2009). Regression analysis showed that honeydew secretion was positively correlated with N, total free sugar and soluble protein contents but negatively correlated with Si content. Relatively less amount of honeydew secretion by BPH was recorded on N deficient. This could be explained in the ways that, the stylet of the BPH might not reach the phloem in those plants containing more Si due to change in plant texture and some chemical reasons (Dale, 1988) or BPH could not suck the phloem sap due to absence of feeding stimulants like soluble proteins and free sugars (Baskaran et al, 1985). Phosphorus addition did not contribute to feeding status BPH as no effect was evident in changing concentrations of nutrients in rice plants except its own level. However, K application reduced the concentrations of total free sugars and soluble proteins in plant tissues, and at the same time, it reduced Si content. This might be the reason why no marked change of honeydew secretion was found by K application. It was reported that soluble sugar and amino acid contents tend to increase in N sufficient and K deficient plants (Marschner, 1995). Nitrogen (including nitrogen in amino acids and proteins) and sugar are two major kinds of herbivore feeding stimulants (Fischer and Fiedler, 2000), while higher silica content and sclerenchymous layer can reduce the feeding rate on different host plants (Dale, 1988).

Insects have intimate and subtle relationships with their host plants which can profoundly affect its suitability to insect pests even minor changes in physiological or chemical attributes of plants. In the present study, duration to cause complete damage of rice plants by BPH infestation was found to decrease with the increase of N level. The increasing susceptibility of rice plants was associated with higher feeding activities of BPH on rice plants with high N rate as this plant contains higher concentrations of tissue N and soluble proteins but lower amount of Si. Heavy application of N fertilizers may be responsible for the heavy crop damage by insects (Lu et al, 2004). Nitrogen has active role in amino acid production and protein synthesis (Sterner and Elser, 2002). As a result concentrations of the primary metabolites amino acids (main nutritional resource of insects) in the plant shoots were increased with increasing N availability (Kajimura et al, 1995; Sauge et al, 2010). Many secondary metabolites produced by plants act as toxins and deterrents for pests and pathogens which are reduced by N (Lou and Baldwin, 2004; Chen and Ni, 2012). However, P fertilization has no effect on tolerance to BPH of rice plants. Although no change in feeding of BPH was evident, K application to rice plants showed increased plant tolerance to the pest which could be for change in some other chemicals like lower levels of soluble proteins and free sugars in plant tissues, thereby creating an unfavorable environment for the insects. It was reported that the levels of soluble sugars, organic acids and amino acids tend to increase in K-deficient plants (Amtmann et al, 2008). Soil K availability and leaf K levels affect plant quality and play an important role in aphid population development (Myers and Gratton, 2006).

RWC is an appropriate estimate of plant water status in terms of cellular water deficit (Lu et al, 2004). In the present study, RWC was significantly higher in N, P and K applied rice plants than those without them. Therefore, applications of N, P and K are found to important in retaining higher water content in rice plants. In this study, RWC decreased drastically in rice plants at the level of 200 kg/hm^2 N with the injury by BPH nymphs compared to those in BPH free plants. On the contrary, RWC in plants without N fertilizer decreased slightly when infested with BPH nymph compared to BPH free plants. RWC of 68.44% in rice plants with 200 kg/hm^2 N supply damaged by 100 nymphs for 5 d reached the wilting scale of RWC (Panda and Khush, 1995). It may be explained that rice plants with higher N content bearing higher water potential can provide planthoppers at low density with enough sap for their higher survival and faster growth. However, at higher density of BPH, RWC of the plants was reduced sharply and the plants became wilted. This might due to the amount of sap sucked by high density BPH exceeded those supplied by rice plants, as there was more feeding rate of sap from host plants with N application. Hence, rice plants with enhanced N and soluble proteins and less effective qualitative defensive chemicals can be more attractive to herbivores, suffering from increased levels of feeding and more plant damage.

ACKNOWLEDGEMENTS

Authors thank M. Mahir UDDIN, Mainul HAQUE, M. Nazmul BARI, M. A. SALEQUE and Rumena YASMEEN for their technical support in conducting the experiments.

REFERENCES

Ali M P, Huang D C, Nachman G, Ahmed N, Begum M A, Rabbi M F. 2014. Will climate change affect outbreak patterns of planthoppers in Bangladesh? *PLoS One*, **9**: e91678.

Altieri M A, Nicholls C I. 2003. Soil fertility management and insect pests: Harmonizing soil and plant health in agroecosystems. *Soil Till Res*, **72**(2): 203–211.

Amtmann A, Troufflard S, Armengaud P. 2008. The effect of potassium nutrition on pest and disease resistance in plants. *Physiol Plant*, **133**(4): 682–691.

Awmack C S, Leather S R. 2002. Host plant quality and fecundity in herbivorous insects. *Annu Rev Entomol*, **47**: 817–844.

Bado S G, Rodriguez S M, Folcia A M. 2002. Variation in abundance of aphids (Homoptera: Aphididae) and predatory ladybirds (Coleoptera: Coccinellidae) in a barley cultivar at different practices of use of fertilizers. *IDESIA*, **20**: 35–42.

Barbour J D, Farrar R R, Kennedy G G. 1991. Interaction of fertilizer regime with host plant resistance in tomato. *Entomol Exp Appl*, **60**(3): 289–300.

Baskaran P, Narayanasamy P, Pari A. 1985. The role of potassium in incidence of insect pests among crop plants, with particular reference to rice. *In*: Role of Potassium in Crop Resistance to Insect Pests. Haryana: Potash Research Institute of India: 63–68.

Bernays E A. 1990. Insect-Plant Interactions. Florida, USA: CRC Press.

Bottrell D G, Schoenly K G. 2012. Resurrecting the ghost of green revolutions past: The brown planthopper as a recurring threat to high-yielding rice production in tropical Asia. *J Asia-Pacific Entomol*, **15**(1): 122–140.

Chen Y, Ni X. 2012. Nitrogen modulation on plant direct and indirect defenses. *In*: Liu T X, Kang L. Recent Advances in Entomological Research: From Molecular Biology to Pest Management. Beijing, China: Higher Education Press: 86–102.

Clark R B. 1982. Plant response to mineral element toxicity and deficiency. *In*: Christiansen M N, Lewis C F. Breeding Plants for Less Favorable Environment. New York, USA: John Wily and Sons: 71–142.

Dale D. 1988. Plant mediated effects of soil mineral stress on insects. *In*: Heinrich E A. Plant Stress Insect Interactions. New York, USA: John Wiley and Sons: 35–110.

Elser J J, Fagan W F, Denno R F, Dobberfuhl D R, Folarin A, Huberty A, Interlandi S, Kilham S S, McCauley E, Schulz K L, Siemann E H, Sterner R W. 2000. Nutritional constraints in terrestrial and freshwater food webs. *Nature*, **408**: 578–580.

Elser J J, Hayakawa K, Urabe J. 2001. Nutrient limitation reduces food quality for zooplankton: *Daphnia* response to seston phosphorus enrichment. *Ecology*, **82**(3): 898–903.

Fagan W F, Siemann E, Denno R F, Mitter C, Huberty A F, Woods H A, Elser J J. 2002. Nitrogen in insects: Implications for trophic complexity and species diversification. *Am Nat*, **160**: 784–802.

Fischer K, Fiedler K. 2000. Response of the copper butterfly *Lycaena tityus* to increased leaf nitrogen in natural food plants: Evidence against the nitrogen limitation hypothesis. *Oecologia*, **124**: 235–241.

Huberty F A, Denno R F. 2006. Consequences of nitrogen and phosphorus limitation for the performance of two planthoppers with divergent life-history strategies. *Oecologia*, **149**(3): 444–455.

International Rice Research Institute (IRRI). 1980. Standard Evaluation System for Rice. Manila, the Phillipines: International Rice Research Institute.

International Rice Research Institute (IRRI). 2007. CROPSTAT for Windows, version 7.2. Manila, the Phillipines: International Rice Research Institute.

Jensen T C, Leinaas H P, Hessen D O. 2006. Age-dependent shift in response to food elemental composition in Collembola: Contrasting effects of dietary nitrogen. *Oecologia*, **149**(4): 583–592.

Kajimura T, Fujisaki K, Nakasuji F. 1995. Effect of organic rice farming on leaffolder and planthoppers: II. Amino acid content in the rice phloem sap and survival rate of planthoppers. *Appl Entomol Zool*, **30**(1): 17–22.

Lou Y, Baldwin I T. 2004. Nitrogen supply influences herbivore-induced direct and indirect defenses and transcriptional responses in *Nicotiana attenuata*. *Plant Physiol*, **135**(1): 496–506.

Lowry O H, Rosenbrough N J, Farr A L, Randall R J. 1951. Protein measurement with the folin phenol reagent. *J Biol Chem*, **193**(1): 265–275.

Lu Z X, Villareal S, Yu X P, Heong K L, Hu C. 2004. Effect of nitrogen on water content, sap flow and tolerance of rice plants to brown planthopper. *Rice Sci*, **11**(3): 129–134.

Lu Z X, Heong K L, Yu X P, Hu C. 2005. Effects of nitrogen nutrient on the behavior of feeding and oviposition of the brown planthopper, *Nilaparvata lugens* on IR64. *J Zhejiang Univ: Agric Life Sci*, **31**: 62–70.

Lu Z X, Heong K L. 2009. Effects of nitrogen-enriched rice plants on ecological fitness of planthoppers. *In*: Heong K L, Hardy B. Planthoppers: New Threats to the Sustainability of Intensive Rice Production Systems in Asia. Manila, the Phillipines: International Rice Research Institute: 247–256.

Marazzi C, Patrian B, Stadler E. 2004. Secondary metabolites of the leaf surface affected by sulphur fertilization and perceived by the diamondback moth. *Chemoecology*, **14**(2): 81–86.

Marschner H. 1995. Mineral Nutrition of Higher Plants. London, UK: Academic Press.

Mongia A D, Singh N T, Mandal L N, Guha A. 1998. Effect of liming, super phosphate and rock phosphate application to rice on the yield and uptake of nutrients on acid sulphate soils. *J Ind Soc Soil Sci*, **46**(1): 61–66.

Moon D C, Stiling P. 2000. Relative importance of abiotically induced direct and indirect effects on a salt-marsh herbivore. *Ecology*, **81**(2): 470–481.

Myers S W, Gratton C. 2006. Influence of potassium fertility on soybean aphid, *Aphis glycines* Matsumura (Hemiptera: Aphididae), population dynamics at a field and regional scale. *Environ Entomol*, **35**(2): 219–227.

Panda N, Khush G S. 1995. Host plant resistance to insects. Wallingford: CAB International.

Pathak P K, Heinrichs E A. 1982. Bromocresol green indicator for measuring feeding activity of *Nilaparvata lugens* on rice varieties. *Phil Entomol*, **5**(2): 209–212.

Prasad B R, Pasalu I C, Raju N B T, Lingaiah T. 2005. Effect of nitrogen levels and rice varieties on brown planthopper adult weight and amount of honeydew excretion. *Ann Plant Prot Sci*, **13**(1): 243–245.

Salim M. 2002a. Effects of potassium nutrition on growth, biomass and chemical composition of rice plants and on host-insect interaction. *Pak J Agric Res*, **17**(1): 14–21.

Salim M. 2002b. Nitrogen induced changes in rice plants: Effect on host-insect interactions. *Pak J Agric Res*, **17**(3): 210–220.

Sarwar M. 2012. Effects of potassium fertilization on population build up of rice stem borers (lepidopteron pests) and rice (*Oryza sativa* L.) yield. *J Cereals Oilseeds*, **3**(1): 6–9.

Sauge M H, Grechil I, Poëssel J L. 2010. Nitrogen fertilization effects on *Myzus persicae* aphid dynamics on peach: Vegetative growth allocation or chemical defense? *Entomol Exp Appl*, **136**(2): 123–133.

Slansky J F, Rodriguez J G. 1987. Nutritional Ecology of Insects, Mites, Spiders and Related Invertebrates. New York: John Wiley & Sons.

Sogawa K, Liu G, Qiang Q. 2009. Prevalence of whitebacked planthoppers in Chinese hybrid rice and whitebacked planthopper resistance in Chinese japonica rice. *In*: Heong K L, Hardy B. Planthoppers: New Threats to the Sustainability of Intensive Rice Production Systems in Asia. Manila, the Phillipines: International Rice Research Institute: 257–280.

SPSS. 2007. SPSS for Windows, version 16. SPSS Inc, Chicago, USA.

Sterner R W, Elser J J. 2002. Ecological Stoichiometry: The Biology of Elements from Molecules to the Biosphere. New Jersey, USA: Princeton University Press.

Stiling P, Moon D C. 2005. Quality or quantity: The direct and indirect effects of host plants on herbivore and their natural enemies. *Oecologia*, **142**(3): 413–420.

Subbaiah P. 1991. Effect of level and source of phosphorus on yield and nutrient uptake of rice (*Oryza sativa* L.). *Ind J Agron*, **36**: 230–232.

Tisdale S L, Nelson W L, Beaton J D. 1985. Soil Fertility and Fertilizers. New York, USA: Macmillan Publication.

Visanuvimol L, Bertram S M. 2011. How dietary phosphorus availability during development influences condition and life history traits of the cricket, *Acheta domesticus*. *J Insect Sci*, **11**: 1–17.

Walker D J, Leigh R A, Miller A J. 1996. Potassium homeostasis in vacuolate plant cells. *Proc Natl Acad Sci USA*, **93**(19): 10510–10514.

Wyn Jones R G, Pollard A. 1983. Proteins, enzymes and inorganic ions. *In*: Lauchli A, Pirson A. Encyclopedia of Plant Physiology. Berlin, Germany: Springer: 528–562.

Wyn Jones R G. 1999. Cytoplasmic potassium homeostasis: Review of the evidence and its implication. *In*: Oosterhuis D M, Berkowitz G A. Frontiers in Potassium Nutrition: New Perspectives on the Effects of Potassium on Physiology of Plants. Atlanta, Georgia, USA: CSSA, Special Publication, Potash and Phosphate Institute: 13–22.

Yoshida S, Forno D A, Cock J H, Gomez K A. 1976. Laboratory Manual for Physiological Studies of Rice. Manila, the Phillipines: International Rice Research Institute.

(Managing Editor: LI Guan)